

## Abstract

Chemical therapy for control and prevention of fish diseases is necessary and a common practice in aquaculture. Many factors affect the accuracy of a chemical treatment application, such as: the functioning of the chemical delivery system, calculation of chemical quantities to be delivered, water temperature, geometry of the culture unit, inlet-outlet structure, influences of aerators, wind movement, and measurement of water volumes and flow rates. Three separate trials were conducted at the Osceola Fish Hatchery (Wisconsin Department of Natural Resources) evaluating the accuracy of flow-through hydrogen peroxide treatments applied to either 1, 3, or 9 raceways that were connected in a series. Raceways were treated with 50 or 75  $\mu\text{L/L}$  of hydrogen peroxide for 30 min. Chemical concentrations were determined titrimetrically. In all applications, the target treatment regimen was not realized. Chemical concentrations dropped and exposure times increased with each additional raceway treated in series. Single introduction of a therapeutic to more than three raceways in series is not recommended. Factors that interfered with the accuracy of the treatments were culture unit configuration, aeration, and flow rates. Several treatment modifications were identified that would result in more accurate chemical treatments.



Osceola Fish Hatchery where chemical trials were conducted

## Objective

To apply a flow-through hydrogen peroxide treatment to either 1, 3, or 9 raceways and analytically verify the accuracy of the treatments. Results from the study will determine if an accurate treatment regimen was applied and may identify factors that adversely affected the application of the chemical treatment.



Hatchery raceways with aeration

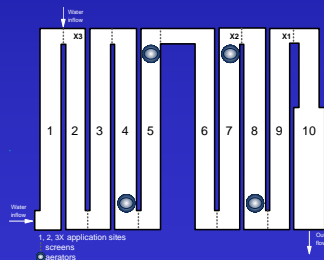


Figure 1.-- Configuration of the 10 raceways at the Osceola Fish Hatchery, Wisconsin. Three trial treatments were conducted; trial 1 raceway 10, trial 2 raceway 8, 9, 10, and trial 3 raceways 2-10. Each treatment was a single point application of hydrogen peroxide.

## Reasons for verifying a chemical treatment

- The expected treatment regimen (exposure time and concentration) must be realized before the effectiveness of a drug can be determined.
- If the expected treatment regimen was not realized - fish culturist must identify and correct factors that were responsible for the inaccurate treatment.
- If corrective measures are unsuccessful in assuring an accurate treatment - the effectiveness of the drug to control a specific fish disease can not be determined.

## Factors that influence the efficacy of a treatment

- Health of the fish
- Identity and severity of the disease
- Environmental conditions
- Accuracy of the treatment application



Technician calibrates pump to deliver chemical stock solution

## Factors Affecting the Accuracy of a Chemical Treatment

- Functioning and calibration of the chemical delivery system
- Measurement of water volumes and flow rates
- Calculation of chemical quantities to be applied
- Degradation of treatment chemical
- Water temperature
- Water quality
- Additional water entering the system (rainwater, run-off, or ground water seepage)
- Geometry of the culture unit (shape, water restrictions, contour)
- Inlet-outlet structure placement
- Physical disturbances of water flow patterns (aerators and wind)
- Physical obstructions (plugged screens)



Mixing of chemical at head of raceway



Chemical was delivered to raceway water at 3 locations

## Methods

- Test article was hydrogen peroxide (35 % active ingredient)
- Hydrogen peroxide treatment regimens were 50 (first trial only) or 75  $\mu\text{L/L}$  applied for 30 min.
- Stock solutions were prepared in 100 L Nalgene tanks.
- Stock solution was applied by drip method
- A peristaltic pump was calibrated to deliver the stock solution (0.93 L/min or 1.37 L/min) to each side and the center area of the incoming water
- Duplicate treatment water samples were collected at mid depth (0.23 m)
- Samples were collected at 5 (Trial 2 only) or 15 min intervals after initiation of the treatment
- Sampling periods; trial 1 (75 min), trial 2 (120 min), and trial 3 (270 min)
- Hydrogen peroxide concentrations were determined titrimetrically

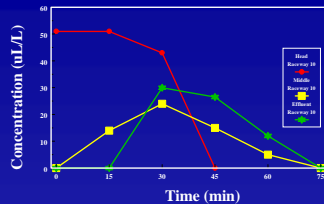


Figure 2.-- Rainbow trout infected with bacterial gill disease were treated with 50  $\mu\text{L/L}$  hydrogen peroxide for 30 min. The hydrogen peroxide concentrations were analytically verified.

In trial 1, rainbow trout in raceway 10 received a flow-through treatment of 50  $\mu\text{L/L}$  for 30 min. The influent end (Figure 2) of the raceway maintained a mean concentration of 48  $\mu\text{L/L}$ , the middle section had a mean concentration of 14.5  $\mu\text{L/L}$ , and the effluent section had a mean concentration of 23  $\mu\text{L/L}$ . There was no detectable hydrogen peroxide in raceway 10 after 75 min. The lower concentration levels recorded at the middle and effluent sections of the raceway were probably the result of the configuration of the raceway which widened in the middle area of the unit. To ensure proper mixing and distribution of the chemical bank within the entire raceway a deflector or aerator should be installed in the middle area of the raceway.

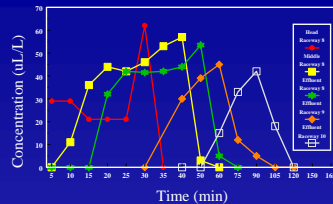


Figure 3.-- Rainbow trout infected with bacterial gill disease were treated with 75  $\mu\text{L/L}$  hydrogen peroxide for 30 min. The hydrogen peroxide concentrations were analytically verified.

In trial 2, rainbow trout in raceways 8, 9, and 10 received a flow-through treatment of 75  $\mu\text{L/L}$  for 30 min. The peak concentrations at the influent, middle, and effluent section of raceway 8 reached 82 %, 76 %, and 72 % of the expected concentration. There were fluctuations in concentration in the influent area of the raceway which were probably the result of adding the chemical near an aerator. This may have drawn part of the chemical bank into the effluent end of raceway 7 or may have caused an eddy effect in this area of the raceway. The chemical bank appeared to be diluted as it reached the effluent end of raceways 9 and 10 with peak concentrations reaching only 60 % and 56 % of the target concentration. There was no detectable hydrogen peroxide in raceway 8, 9, or 10 after 75, 105, and 120 min. The information indicated three raceways could be treated in series. However in this particular treatment, personnel would have to determine the cause of the lower treatment concentration and then correct the problem to ensure accurate chemical treatments in the future.

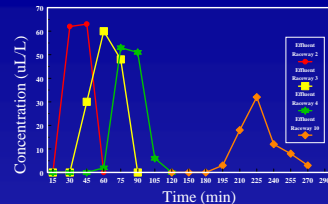


Figure 4.-- Rainbow trout infected with bacterial gill disease were treated with 75  $\mu\text{L/L}$  hydrogen peroxide for 30 min. The hydrogen peroxide concentrations were analytically verified.

In trial 3, rainbow trout held in a series of 9 raceways (raceways 2-10) received a flow-through treatment of 75  $\mu\text{L/L}$  for 30 min. Results indicated the peak concentration in the effluent area of raceway 2 reached approximately 84 % of the expected concentration and the concentration was not maintained for the entire 30 min exposure period. The chemical bank was apparently diluted as it reached the effluent end of raceways 3 and 4 where peak concentrations (maintained for <15 min) reached only 64 % and 71 % of the target concentration, respectively. The concentration of the chemical bank was reduced to 43 % of the expected concentration when the chemical bank reached the effluent end of raceway 10, and hydrogen peroxide was detected at that sampling location for over 90 min. There was no detectable hydrogen peroxide in raceway 2, 3, 4 or 10 after 60, 90, 120, and 270 min, respectively. The only way to treat an extended number of raceways (9 in this example) is to boost the chemical bank (add additional chemical) as it passes specific raceways.

## Conclusions

- This study provided information on procedures that could be implemented to verify the accuracy of a chemical treatment regimen.
- Procedures developed should apply to verification of most chemical treatment regimens (flowing or static treatments) at any fish rearing facility.
- If an inaccurate treatment occurs, fish culturists should conduct hydrological tests to identify the variables negatively affecting the treatment.
- Fish culturists should implement corrective measures to ensure future treatments are accurate.
- Implementation of procedures to analytically verify waterborne drug applications should result in accurate and consistent treatments.

